

DECREASED RATE OF CORONARY RESTENOSIS AFTER LOWERING OF PLASMA HOMOCYSTEINE LEVELS

GUIDO SCHNYDER, M.D., MARCO ROFFI, M.D., RICCARDO PIN, M.D., YVONNE FLAMMER, M.D., HELMUT LANGE, M.D., FRANZ R. EBERLI, M.D., BERNHARD MEIER, M.D., ZOLTAN G. TURI, M.D., AND OTTO M. HESS, M.D.

ABSTRACT

Background We have previously demonstrated an association between elevated total plasma homocysteine levels and restenosis after percutaneous coronary angioplasty. We designed this study to evaluate the effect of lowering plasma homocysteine levels on restenosis after coronary angioplasty.

Methods A combination of folic acid (1 mg), vitamin B₁₂ (400 µg), and pyridoxine (10 mg) — referred to as folate treatment — or placebo was administered to 205 patients (mean [±SD] age, 61±11 years) for six months after successful coronary angioplasty in a prospective, double-blind, randomized trial. The primary end point was restenosis within six months as assessed by quantitative coronary angiography. The secondary end point was a composite of major adverse cardiac events.

Results Base-line characteristics and initial angiographic results after coronary angioplasty were similar in the two study groups. Folate treatment significantly lowered plasma homocysteine levels from 11.1±4.3 to 7.2±2.4 µmol per liter (P<0.001). At follow-up, the minimal luminal diameter was significantly larger in the group assigned to folate treatment (1.72±0.76 vs. 1.45±0.88 mm, P=0.02), and the degree of stenosis was less severe (39.9±20.3 percent vs. 48.2±28.3 percent, P=0.01). The rate of restenosis was significantly lower in patients assigned to folate treatment (19.6 percent vs. 37.6 percent, P=0.01), as was the need for revascularization of the target lesion (10.8 percent vs. 22.3 percent, P=0.047).

Conclusions Treatment with a combination of folic acid, vitamin B₁₂, and pyridoxine significantly reduces homocysteine levels and decreases the rate of restenosis and the need for revascularization of the target lesion after coronary angioplasty. This inexpensive treatment, which has minimal side effects, should be considered as adjunctive therapy for patients undergoing coronary angioplasty. (N Engl J Med 2001;345:1593-600.)

Copyright © 2001 Massachusetts Medical Society.

THE occurrence of restenosis after percutaneous coronary angioplasty remains an important limitation of the procedure,¹ and effective pharmacotherapy has been elusive.²⁻⁵ Thus, the observation that the total plasma homocysteine level is an important predictor of cardiovascular risk^{6,7} and correlates with the severity of coronary artery disease^{8,9} has led to interest in its potential role in restenosis. Although the mechanism of homocysteine-induced vascular damage is not known, a number of

potential links have been suggested.¹⁰⁻¹⁴ We have previously shown that patients with plasma homocysteine levels below 9 µmol per liter have a 49 percent lower rate of coronary restenosis than patients with higher plasma homocysteine levels.¹⁵ Since plasma homocysteine can be reliably lowered 25 to 30 percent with a daily dose of at least 500 µg of folic acid in combination with vitamin B₁₂ and pyridoxine,^{6,16} we hypothesized that lowering of homocysteine levels would decrease the rate of restenosis after coronary angioplasty.

METHODS**Study Design**

We conducted a prospective, double-blind, randomized trial, enrolling consecutive patients who had undergone successful angioplasty of at least one coronary stenosis of 50 percent or more. The study protocol was approved by the local ethics committee, and patients gave written informed consent. Patients who had unstable angina, myocardial infarction within the previous two weeks, clinically significant disease of the left main artery, angioplasty of a bypassed vessel with a patent graft, or renal dysfunction (defined by a serum creatinine level of more than 1.8 mg per deciliter [160 µmol per liter]) or who were taking multivitamins, participating in other trials, or unwilling to undergo follow-up angiography were not enrolled. Patients were randomly assigned to receive either folic acid (1 mg), vitamin B₁₂ (400 µg), and pyridoxine (10 mg) daily — subsequently referred to as folate treatment — or placebo. Fasting levels of total plasma homocysteine were measured at admission and at follow-up examination with the use of the technique described by Ubbink et al., a sensitive and reproducible method with a coefficient of variation of 6.6 percent and a lower limit of detection of 2 µmol per liter.¹⁷

Percutaneous Coronary Angioplasty

Coronary angioplasty was performed with standard guide wires and balloon catheters. The pressure and duration of inflation, as well as the use of stents and adjunctive drug therapy (heparin, aspirin, ticlopidine or clopidogrel, or glycoprotein IIb/IIIa inhibitors), were left to the discretion of the operator. Successful coronary angioplasty was defined as residual stenosis of less than 35 percent with a normal (Thrombolysis in Myocardial Infarction grade 3) flow pattern. Clinical and angiographic follow-up was performed at six months, or earlier if symptoms recurred. Follow-up angiographic data obtained less than three months after coronary angioplasty were included if restenosis was documented; otherwise, patients were asked to return for the six-month follow-up examination.

Angiographic Evaluation

Base-line coronary angiograms were obtained in two orthogonal views after dilation with nitrates. Quantitative coronary angiography

From the Division of Cardiology, Swiss Cardiovascular Center Bern, University Hospital, Bern, Switzerland (G.S., M.R., R.P., Y.F., F.R.E., B.M., O.M.H.); the Kardiologische Praxis, Bremen, Germany (H.L.); and the Division of Cardiology, University of California at San Diego Medical Center, San Diego (Z.G.T.). Address reprint requests to Dr. Schnyder at the University of California at San Diego Medical Center, Cardiology Division, 200 W. Arbor Dr., San Diego, CA 92103-8784, or at g.schnyder@lycos.com.

was performed with the use of an automated edge-detection system (Philips Integris-BH-3000, Version 2, if on-line, or Philips ViewStation-CDM-3500, Version 2, if off-line; Philips, Best, the Netherlands). The tip of the diagnostic or guiding catheter (positioned at the coronary ostium) was used as a scaling device to obtain absolute arterial dimensions. The same views and calibration techniques were used at follow-up examination. End-diastolic frames in the two orthogonal views showing maximal severity of stenosis were chosen for measurement of the luminal diameter. The reference diameter of the vessel, the minimal diameter of the lumen, the degree of stenosis — expressed as a percentage of the diameter of the vessel — and the length of the lesion were calculated as the average value of the two views. Late loss in luminal diameter was defined as the minimal luminal diameter immediately after coronary angioplasty minus the minimal luminal diameter at follow-up. Restenosis was defined as stenosis of 50 percent or more at follow-up examination. Patients who had more than one lesion treated were defined as having restenosis if at least one dilated artery fulfilled the criteria for restenosis. Angiograms were analyzed by an experienced interventional cardiologist who was unaware of homocysteine levels or treatment assignments. The intraobserver variability for minimal luminal diameter and degree of stenosis was 0.15 ± 0.22 mm and 7 ± 12 percent, respectively.

Study End Points

The primary end point with respect to efficacy was the presence or absence of restenosis of 50 percent or more at follow-up examination. An additional analysis of the rate of restenosis per dilated lesion was performed. The rate of restenosis was also analyzed according to the absolute and relative reduction in homocysteine levels achieved. The secondary end point was a composite of major adverse cardiac events defined as death from cardiac causes, nonfatal myocardial infarction (new pathologic Q waves), or revascularization of the target lesion.

Statistical Analysis

A sample size of 91 patients in each treatment group was needed to achieve a statistical power of 0.80 to detect a 20 percent reduction in the absolute rate of restenosis. To account for the possibility of patients lost to follow-up, the planned sample size was 205 patients. Skewed variables were log-transformed before analysis. Results are shown in natural units. Categorical variables are reported as counts (percentages) and continuous variables as means \pm SD. For categorical variables, a continuity-corrected chi-square test was used to test differences between the two treatment groups. For continuous variables, a two-tailed t-test was employed. The Spearman rank-correlation coefficient was used to estimate the correlation between late loss of luminal diameter and homocysteine levels at follow-up. Kaplan–Meier survival curves were used to evaluate freedom from major adverse cardiac events, and differences in the treatment effect were assessed with the Mantel–Cox log-rank test. Multiple logistic-regression analysis was used to evaluate the relation between angiographically identified restenosis and multiple clinical and angiographic variables, including the use of stents, the treatment of restenotic lesions, the size of the vessels involved, the postprocedural minimal luminal diameter, and the location of the target lesion. A two-tailed P value of less than 0.05 was considered to indicate statistical significance. Data were prospectively collected and analyzed with the use of StatView software (version 4.5, SAS Institute, Cary, N.C.).

RESULTS

A total of 205 patients were randomly assigned to either folate treatment (105 patients) or placebo (100 patients). Twenty-eight patients did not complete follow-up: 9 (3 assigned to folate treatment and 6 to placebo) discontinued the study medication and declined clinical follow-up and follow-up angiography, 16 (9 as-

signed to folate treatment and 7 to placebo) declined follow-up angiography, and 3 (1 assigned to folate treatment and 2 to placebo) died before the follow-up reevaluation. This left a total of 196 patients (95.6 percent) with clinical follow-up data and 177 patients (86.3 percent) with angiographic follow-up data. One patient assigned to folate treatment discontinued the study medication because of pruritus. No other side effect was reported. In terms of base-line clinical, laboratory, and angiographic criteria, the 28 patients without angiographic follow-up data and the 9 patients without clinical follow-up data did not differ significantly from the remaining population.

Clinical Characteristics and Laboratory Findings

The two study groups were similar in terms of sex, age, and cardiovascular risk factors (Table 1). Twenty-two percent of the patients were women; the mean age was 61 years, and the distribution of cardiovascular risk factors was typical of a population in central Europe. The base-line demographic characteristics, the severity of coronary artery disease (as measured by the presence or absence of a history of previous myocardial infarction, the prior use or nonuse of revascularization with bypass surgery or angioplasty, and the number of treated lesions per patient), and the base-line laboratory values were not significantly different between the two study groups. As expected, homocysteine levels at follow-up were significantly lower in patients assigned to folate treatment than in those assigned to placebo (7.2 ± 2.4 vs. 9.5 ± 3.6 μ mol per liter, $P < 0.001$).

Angiographic Analysis

The mean duration of angiographic follow-up was 27 ± 6 weeks. There was no significant difference between the two study groups with regard to the size of the vessels involved, the minimal luminal diameter, and the degree of stenosis before and immediately after coronary angioplasty (Table 2). There was a somewhat higher rate of use of stents in control patients ($P = 0.23$), whereas the rate of use of glycoprotein IIb/IIIa inhibitors was similar in the two groups. At follow-up, lesions in the group assigned to folate treatment had a larger minimal luminal diameter (1.72 ± 0.76 vs. 1.45 ± 0.88 mm, $P = 0.02$) and less severe stenosis (39.9 ± 20.3 percent vs. 48.2 ± 28.3 percent, $P = 0.01$). In Figure 1, the minimal luminal diameters before and immediately after coronary angioplasty demonstrate the similarity of the two study groups at base line and the similar angiographic gain after angioplasty. However, at follow-up a higher amount of late loss of luminal diameter can be seen in the control group (0.82 ± 0.76 vs. 0.61 ± 0.74 mm, $P = 0.03$). There was a correlation between late loss of luminal diameter and homocysteine levels at follow-up ($r = 0.27$, $P < 0.001$; a loss of 0.1 mm of luminal diameter per 1.7 μ mol of plasma homocysteine per li-

TABLE 1. CLINICAL CHARACTERISTICS AND LABORATORY FINDINGS.*

VARIABLE	FOLATE TREATMENT (N=105)	CONTROL (N=100)	P VALUE
Sex (M/F)	80/25	79/21	0.76
Age (yr)	61.3±11.3	61.1±11.5	0.91
Smoker (%)†	40	43	0.77
Diabetes mellitus (%)‡	26	28	0.84
Arterial hypertension (%)§	62	65	0.76
Hypercholesterolemia (%)¶	85	78	0.26
Previous myocardial infarction (%)	57	57	0.90
Myocardial infarction within the previous six months (%)	31	33	0.93
Previous percutaneous transluminal coronary angioplasty (%)	29	30	0.95
Previous coronary-artery bypass grafting (%)	11	12	0.93
Number of treated lesions	1.35±0.65	1.31±0.61	0.67
Laboratory findings			
Glycosylated hemoglobin (%)	6.0±1.2	6.0±1.0	0.81
Creatinine (mg/dl)	1.04±0.22	1.04±0.18	0.99
Homocysteine (μmol/liter)			
Base line	11.1±4.3	10.8±4.0	0.61
Follow-up	7.2±2.4	9.5±3.6	<0.001
Cholesterol (mg/dl)	216±46	212±46	0.84
High-density lipoprotein cholesterol (mg/dl)	46±12	46±15	0.76
Low-density lipoprotein cholesterol (mg/dl)	127±39	131±46	0.73
Triglycerides (mg/dl)	195±142	177±133	0.51
Therapy at discharge (%)			
Statins	72	70	0.83
Beta-blockers	63	69	0.44
Angiotensin-converting-enzyme inhibitors	35	34	0.97

*Plus-minus values are means ±SD. To convert values for cholesterol to millimoles per liter, multiply by 0.0259; to convert values for triglycerides to millimoles per liter, multiply by 0.0113; to convert values for creatinine to micromoles per liter, multiply by 88.4.

†Smokers were defined as current smokers and patients who had discontinued smoking within the previous six months.

‡Diabetes mellitus was defined by a glycosylated hemoglobin level of at least 6.2 percent or the current use of insulin or oral hypoglycemic therapy.

§Hypertension was defined by an arterial pressure of at least 140/90 mm Hg or the current use of antihypertensive therapy.

¶Hypercholesterolemia was defined by a cholesterol level of at least 200 mg per deciliter (5.2 mmol per liter) or the current use of lipid-lowering drugs.

||The normal range of plasma homocysteine levels is considered to be between 5 and 15 μmol per liter, but many authors believe that the upper limit of the normal range should be 10 to 12 μmol per liter.^{7,8,18}

ter). This correlation was stronger for lesions treated with balloon angioplasty only ($r=0.48$, $P<0.001$; 0.1-mm loss of luminal diameter per 1.2 μmol of plasma homocysteine per liter). This correlation was not reproducible for stented lesions ($r=0.07$, $P=0.44$).

End Points

In the group assigned to folate treatment, 19.6 percent (18 of 92 patients) reached the primary end point of restenosis, as compared with 37.6 percent (32 of 85) in the control group ($P=0.01$), corresponding to a relative reduction of 48 percent (relative risk, 0.52; 95 percent confidence interval, 0.32 to 0.86). When individual lesions were considered, there was a 15.7 percent (19 of 121 lesions) rate of restenosis in the group assigned to folate treatment, as compared with

a 34.5 percent (38 of 110 lesions) rate of restenosis in the control group ($P=0.002$), corresponding to a relative reduction of 54 percent (relative risk, 0.46; 95 percent confidence interval, 0.28 to 0.73) (Fig. 2). In 101 lesions treated with balloon angioplasty only, there was a relative reduction of 76 percent with folate treatment (10.3 percent [6 of 58 lesions] vs. 41.9 percent [18 of 43 lesions], $P<0.001$; relative risk, 0.25; 95 percent confidence interval, 0.11 to 0.57). In 130 stented lesions, we observed a trend toward a lower rate of restenosis with folate treatment (20.6 percent [13 of 63 lesions] vs. 29.9 percent [20 of 67 lesions], $P=0.32$; relative risk, 0.69; 95 percent confidence interval, 0.38 to 1.27). The absolute and relative reductions in homocysteine levels were greater in patients without restenosis than in those with resteno-

TABLE 2. CHARACTERISTICS OF 231 LESIONS WITH ANGIOGRAPHIC FOLLOW-UP, AND TREATMENT OPTIONS.*

CHARACTERISTIC	FOLATE TREATMENT (N=121)	CONTROL (N=110)	P VALUE
Lesion location (%)†			
Left anterior descending coronary artery	48	54	0.49
Proximal left anterior descending coronary artery	18	20	0.86
Circumflex coronary artery	23	24	0.95
Right coronary artery	29	23	0.36
Restenotic lesions (%)	7	6	0.86
Complex lesions (%)‡	61	63	0.92
Treatment options (%)			
Stenting	52	61	0.23
Glycoprotein IIb/IIIa inhibitors	12	14	0.94
Plasma homocysteine ($\mu\text{mol/liter}$)			
Base line	11.0 \pm 3.9	10.8 \pm 3.9	0.71
Follow-up	7.3 \pm 2.4	9.3 \pm 3.6	<0.001
Reference-vessel diameter (mm)			
Before angioplasty	2.84 \pm 0.69	2.83 \pm 0.63	0.90
After angioplasty	3.05 \pm 0.68	2.95 \pm 0.58	0.26
At follow-up	2.83 \pm 0.68	2.72 \pm 0.54	0.19
Minimal luminal diameter (mm)			
Before angioplasty	0.95 \pm 0.49	0.86 \pm 0.47	0.16
After angioplasty	2.33 \pm 0.64	2.27 \pm 0.57	0.45
At follow-up	1.72 \pm 0.76	1.45 \pm 0.88	0.02
Degree of stenosis (%)			
Before angioplasty	66.8 \pm 15.1	69.2 \pm 15.6	0.24
After angioplasty	23.6 \pm 10.1	23.4 \pm 10.1	0.85
At follow-up	39.9 \pm 20.3	48.2 \pm 28.3	0.01
Lesion length (mm)	12.7 \pm 7.5	12.5 \pm 6.3	0.84

*Plus–minus values are means \pm SD.

†Because of rounding, not all values sum to 100.

‡Complex lesions were defined as lesions of types B2 and C according to the modified classification of the American College of Cardiology and the American Heart Association.¹⁹

sis (3.1 \pm 3.6 vs. 1.8 \pm 3.9 μmol per liter, $P=0.037$, and 26.6 \pm 37.9 percent vs. 12.5 \pm 43.1 percent, $P=0.038$, respectively). Finally, among patients treated with folate, 14.1 percent (13 of 92 patients) had no response, with homocysteine levels at follow-up unchanged or higher than at the time they entered the study. In these patients, folate treatment did not provide any significant improvement in the rate of restenosis as compared with control patients (30.8 percent [4 of 13 patients] vs. 37.6 percent [32 of 85 controls], $P=0.87$).

Multivariate analysis including variables known to influence restenosis after coronary angioplasty (the use of stents, the treatment of restenotic lesions, the size of the vessels involved, the postprocedural minimal luminal diameter, and the location of the target lesion) did not significantly change the ability of folate treatment to lower the rate of restenosis after coronary angioplasty. After multivariate analysis, only folate treatment ($P=0.007$) and prior restenosis ($P=0.011$) retained significance.

There was a lower incidence of major adverse cardiac events at six months in patients assigned to folate

treatment (12.7 percent [13 of 102 patients]) than in control patients (24.5 percent [23 of 94 controls], $P=0.055$; relative risk, 0.52; 95 percent confidence interval, 0.28 to 0.98). When analyzed at each time point during the follow-up period, this difference between treatment groups is significant ($P=0.02$) (Fig. 3). This difference in the composite end point was primarily due to a reduced rate of revascularization of the target lesion (10.8 percent [11 of 102 patients] vs. 22.3 percent [21 of 94 controls], $P=0.047$; relative risk, 0.48; 95 percent confidence interval, 0.25 to 0.94). No difference was seen between the two groups in the rate of death from cardiac causes (1.0 percent [1 of 102 patients] vs. 2.1 percent [2 of 94 controls], $P=0.95$) and nonfatal myocardial infarction (4.9 percent [5 of 102 patients] vs. 7.4 percent [7 of 94 controls], $P=0.66$).

DISCUSSION

Homocysteine levels are modulated through a series of steps in the pyridoxal phosphate–dependent cystathionine β -synthase pathway or through vitamin B₁₂– and folate-dependent remethylation to methionine.

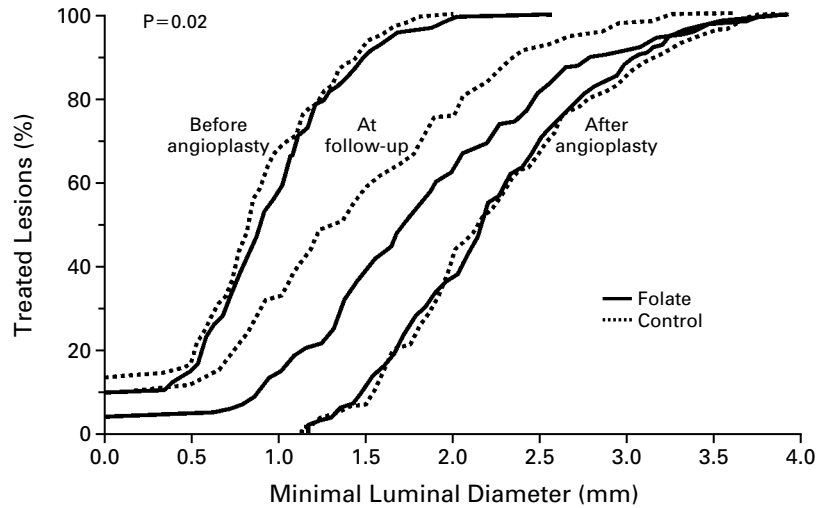


Figure 1. Cumulative Distribution of Minimal Luminal Diameters at Base Line, Immediately after Percutaneous Coronary Angioplasty, and at Six Months of Follow-up for 231 Lesions. The curves for the minimal luminal diameter at base line (before coronary angioplasty) and after coronary angioplasty are nearly identical in the two groups, confirming similarity at base line and similar angiographic gain. However, the curves for the minimal luminal diameter at six months show more late loss of luminal diameter in lesions in the control group. P=0.02 for the comparison of minimal luminal diameters at follow-up in the two groups.

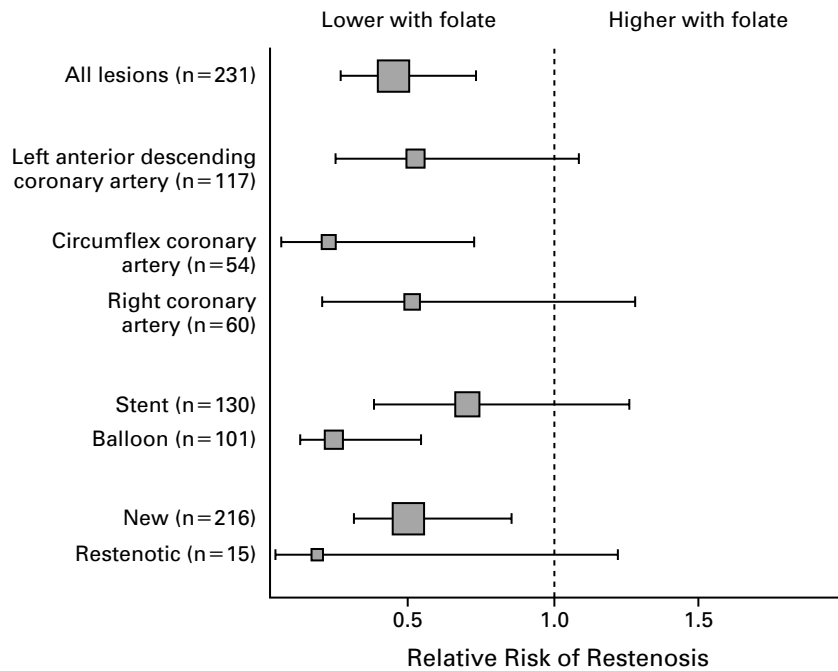


Figure 2. Risk of Restenosis with Folate Treatment among the Total Study Population and Subgroups Stratified According to the Location of the Vessel Involved, the Use or Nonuse of Stents in the Lesions, and the Type of Lesion (New or Restenotic). Squares indicate the relative risk of restenosis in the group assigned to folate treatment as compared with the control group; the size of each square is proportional to the number of lesions, and the horizontal bars represent 95 percent confidence intervals.

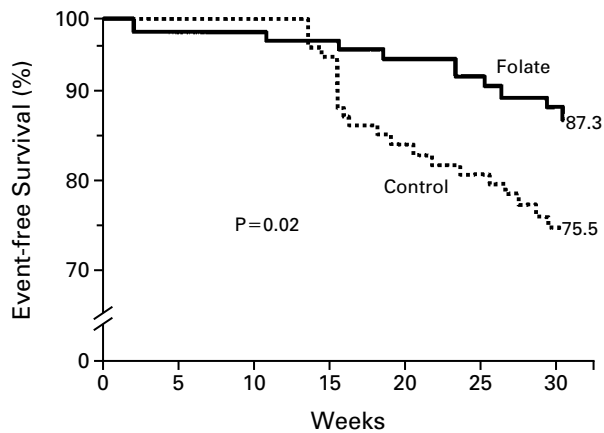


Figure 3. Kaplan–Meier Analysis of Freedom from Major Adverse Cardiac Events in 196 Patients.

The rate of event-free survival was significantly higher among patients assigned to folate treatment than among control patients. The relative risk of a major cardiac event with folate treatment was 0.52 (95 percent confidence interval, 0.28 to 0.98). Revascularization of the target lesion (relative risk, 0.48; 95 percent confidence interval, 0.25 to 0.94) accounted for most of the observed events.

It has been suggested that partial deficiencies of cystathionine β -synthase or 5',10'-methylene-tetrahydrofolate reductase are associated with mild-to-moderate elevations of plasma homocysteine levels and lead to vascular disease.²⁰ Elevated homocysteine levels may reflect either genetic defects (in up to 14 percent of patients)^{20,21} or acquired conditions such as folate, pyridoxine, and vitamin B₁₂ deficiencies or renal failure. On the basis of our previous findings showing that moderately elevated homocysteine levels are associated with restenosis after coronary angioplasty,¹⁵ the present study was designed to evaluate the effect of the lowering of homocysteine levels on the rate of restenosis.

The study provides evidence that folate treatment lowers plasma homocysteine levels, significantly reduces the rate of restenosis after coronary angioplasty, and — primarily through a reduction in the rate of revascularization of the target lesion — decreases the incidence of major adverse cardiac events. These results were obtained with minimal side effects and at a very low cost. Although other therapeutic approaches, such as radiation therapy, have been proposed to achieve similar results,²² the low cost and risk–benefit ratio of folate therapy is appealing. The lowering of plasma homocysteine levels was of particular benefit in non-stented lesions, potentially challenging the current trend of primary stenting. The size of the vessels involved, the postprocedural minimal luminal diameter, the treatment of restenotic lesions, and the location of the target lesion have been shown to influence the rate of restenosis.^{23,24} These variables were equally dis-

tributed between the two study groups, and the power of folate treatment to lower the rate of restenosis remained unaltered in multivariate analysis. The rate of restenosis of 37.6 percent in control patients reflects our relatively broad criteria for inclusion, including lesions in small vessels (less than 3.0 mm in diameter) and segments previously treated for restenosis, both of which have a high risk of restenosis (40 percent²⁵ and more than 50 percent,²⁶ respectively). However, these characteristics were equally distributed between the two study groups and therefore did not influence our findings.

The pathogenesis of homocysteine-induced vascular damage and its possible role in restenosis are not clearly understood. Nevertheless, several hypotheses have been suggested. Elevated homocysteine levels stimulate proliferation of vascular smooth-muscle cells,^{10,11} increase collagen deposition,²⁷ impair endothelium-dependent vasodilation,¹² promote intimal thickening,¹³ and increase the production of extracellular superoxide dismutase.¹⁴ There is also a clear association between elevated homocysteine levels and increased thrombogenicity through interaction with coagulation factor V,²⁸ protein C,²⁹ tissue plasminogen activator,³⁰ and tissue factor activity.³¹ In a manner analogous to the potent antioxidant properties of probucol,³ the oxidant properties¹⁴ of homocysteine may also influence the occurrence of restenosis, even though other antioxidants — i.e., beta carotene, vitamin E, and vitamin C — have failed to reduce the rate of restenosis after coronary angioplasty.³

Since the reduction in the rate of restenosis with folate treatment was greatest in the lesions treated with only balloon angioplasty, one may postulate a positive effect on both vascular remodeling and neointimal hyperplasia. However, since the method of treatment (stent or balloon only) was left to the discretion of the operator, these two subgroups cannot be readily compared, even though the base-line characteristics of the lesions were similar, with the exception of 15 restenotic lesions, which were all treated with stents ($P=0.001$). Furthermore, because intravascular ultrasonography was not performed, the issue of the pathophysiologic mechanism cannot be definitively addressed.

A critical question is whether the strong association between the lowering of plasma homocysteine levels and the decrease in the rate of restenosis reflects causality. Even though the two study groups were similar in terms of base-line clinical, laboratory, and angiographic criteria, there was a nonsignificant trend toward a greater use of stents in the control group; the use of stents has been shown to reduce the rate of restenosis.^{32,33} Despite this, the rate of restenosis was higher in the control group. It could be speculated that the higher rate of restenosis reflects the presence of more complex lesions in the control group. This possibility cannot be excluded, but the similar severity and morphologic features of the lesions¹⁹ at base

line as well as the similar angiographic results after angioplasty between the two study groups do not support it. Thus, the trend toward a lower use of stents in the group assigned to folate treatment may be seen to strengthen our findings indirectly.

Other potential limitations merit consideration. We cannot be certain whether the benefit seen was due solely to lower homocysteine levels or was also influenced by other effects of folate treatment. Despite the findings of the Homocysteine Lowering Trialists' Collaboration that pyridoxine does not significantly lower homocysteine levels,¹⁶ pyridoxine has a number of important actions. Pyridoxine deficiency appears to be an independent predictor for coronary artery disease³⁴ and has been shown to alter platelet function.³⁵ The administration of pyridoxine to the group assigned to folate treatment, or possibly another effect of folate treatment unrelated to homocysteine, could have contributed to the improvement seen with folate therapy. Nevertheless, our previous work showing a direct association between homocysteine levels and restenosis after coronary angioplasty,¹⁵ as well as the present findings (a correlation between late loss of luminal diameter and homocysteine levels at follow-up, an absence of significant benefit in patients with no lowering of homocysteine levels among those assigned to folate treatment, and a significantly greater absolute and relative reduction in homocysteine levels in patients free of restenosis), all support the hypothesis that folate treatment, which lowers homocysteine levels, is effective in preventing restenosis in patients undergoing coronary angioplasty.

Supported by a career development grant from the Swiss National Science Foundation (to Dr. Schnyder) and by the University Hospital, Bern, Switzerland.

We are indebted to the patients and their physicians for their participation in this study and to the staff of the Coronary Catheterization Laboratory and the nursing staff of the Swiss Cardiovascular Center in Bern for their cooperation.

REFERENCES

- Serruys PW, Luijten HE, Beatt KJ, et al. Incidence of restenosis after successful coronary angioplasty: a time-related phenomenon: a quantitative angiographic study in 342 consecutive patients at 1, 2, 3, and 4 months. *Circulation* 1988;77:361-71.
- The EPILOG Investigators. Platelet glycoprotein IIb/IIIa receptor blockade and low-dose heparin during percutaneous coronary revascularization. *N Engl J Med* 1997;336:1689-96.
- Tardif J-C, Côté G, Lespérance J, et al. Probulcol and multivitamins in the prevention of restenosis after coronary angioplasty. *N Engl J Med* 1997;337:365-72.
- Tsuchikane E, Fukuhara A, Kobayashi T, et al. Impact of cilostazol on restenosis after percutaneous coronary balloon angioplasty. *Circulation* 1999;100:21-6.
- Kiesz RS, Buszman P, Martin JL, et al. Local delivery of enoxaparin to decrease restenosis after stenting: results of initial multicenter trial: Polish-American Local Lovenox NIR Assessment study (the POLONIA study). *Circulation* 2001;103:26-31.
- Boushey CJ, Beresford SAA, Omenn GS, Motulsky AG. A quantitative assessment of plasma homocysteine as a risk factor for vascular disease: probable benefits of increasing folic acid intakes. *JAMA* 1995;274:1049-57.
- Seshadri N, Robinson K. Homocysteine, B vitamins, and coronary artery disease. *Med Clin North Am* 2000;84:215-37.
- Chao CL, Tsai HH, Lee CM, et al. The graded effect of hyperhomocysteinemia on the severity and extent of coronary atherosclerosis. *Atherosclerosis* 1999;147:379-86.
- Schnyder G, Pin R, Roffi M, Flammer Y, Hess OM. Association of plasma homocysteine with the number of major coronary arteries severely narrowed. *Am J Cardiol* 2001;88:1027-30.
- Tsai JC, Perrella MA, Yoshizumi M, et al. Promotion of vascular smooth muscle cell growth by homocysteine: a link to atherosclerosis. *Proc Natl Acad Sci U S A* 1994;91:6369-73.
- Tang L, Mamotte CD, Van Bockxmeer FM, Taylor RR. The effect of homocysteine on DNA synthesis in cultured human vascular smooth muscle. *Atherosclerosis* 1998;136:169-73.
- Tawakol A, Omland T, Gerhard M, Wu JT, Creager MA. Hyperhomocyst(e)inemia is associated with impaired endothelium-dependent vasodilation in humans. *Circulation* 1997;95:1119-21. [Erratum, *Circulation* 2000;101:E116.]
- Starkebaum G, Harlan JM. Endothelial cell injury due to copper-catalyzed hydrogen peroxide generation from homocysteine. *J Clin Invest* 1986;77:1370-6.
- Wang XL, Duarte N, Cai H, et al. Relationship between total plasma homocysteine, polymorphisms of homocysteine metabolism related enzymes, risk factors and coronary artery disease in the Australian hospital-based population. *Atherosclerosis* 1999;146:133-40.
- Schnyder G, Roffi M, Flammer Y, Pin R, Hess O. Association of plasma homocysteine with restenosis after percutaneous coronary angioplasty. *Eur Heart J* (in press).
- Homocysteine Lowering Trialists' Collaboration. Lowering blood homocysteine with folic acid based supplements: meta-analysis of randomised trials. *BMJ* 1998;316:894-8.
- Ubbink JB, Hayward-Vermaak WJ, Bissbort S. Rapid high-performance liquid chromatographic assay for total homocysteine levels in human serum. *J Chromatogr* 1991;565:441-6.
- Selhub J, Jacques PF, Bostom AG, et al. Association between plasma homocysteine concentrations and extracranial carotid-artery stenosis. *N Engl J Med* 1995;332:286-91.
- Ellis SG, Vandormael MG, Cowley MJ, et al. Coronary morphologic and clinical determinants of procedural outcome with angioplasty for multivessel coronary disease: implications for patient selection. *Circulation* 1990;82:1193-202.
- Genest JJ Jr, McNamara JR, Upson B, et al. Prevalence of familial hyperhomocyst(e)inemia in men with premature coronary artery disease. *Arterioscler Thromb* 1991;11:1129-36.
- Brattstrom L, Wilcken DEL, Ohrvik J, Brudin L. Common methylenetetrahydrofolate reductase gene mutation leads to hyperhomocysteinemia but not to vascular disease: the result of a meta-analysis. *Circulation* 1998;98:2520-6.
- Raizner AE, Oesterle SN, Waksman R, et al. Inhibition of restenosis with beta-emitting radiotherapy: report of the Proliferation Reduction with Vascular Energy Trial (PREVENT). *Circulation* 2000;102:951-8.
- Hirshfeld JW Jr, Schwartz JS, Jugo R, et al. Restenosis after coronary angioplasty: a multivariate statistical model to relate lesion and procedure variables to restenosis. *J Am Coll Cardiol* 1991;18:647-56.
- Ellis SG, Cowley MJ, DiSciascio G, et al. Determinants of 2-year outcome after coronary angioplasty in patients with multivessel disease on the basis of comprehensive preprocedural evaluation: implications for patient selection. *Circulation* 1991;83:1905-14.
- Briguori C, Nishida T, Adamian M, et al. Coronary stenting versus balloon angioplasty in small coronary artery with complex lesions. *Catheter Cardiovasc Interv* 2000;50:390-7.
- Teirstein PS, Massullo V, Jani S, et al. Catheter-based radiotherapy to inhibit restenosis after coronary stenting. *N Engl J Med* 1997;336:1697-703.
- Majors A, Ehrhart LA, Pezacka EH. Homocysteine as a risk factor for vascular disease: enhanced collagen production and accumulation by smooth muscle cells. *Arterioscler Thromb Vasc Biol* 1997;17:2074-81.
- Rodgers GM, Kane WH. Activation of endogenous factor V by a homocysteine-induced vascular endothelial cell activator. *J Clin Invest* 1986;77:1909-16.
- Rodgers GM, Conn MT. Homocysteine, an atherogenic stimulus, reduces protein C activation by arterial and venous endothelial cells. *Blood* 1990;75:895-901.
- Hajjar KA, Mauri L, Jacovina AT, et al. Tissue plasminogen activator binding to the annexin II tail domain: direct modulation by homocysteine. *J Biol Chem* 1998;273:9987-93.
- Fryer RH, Wilson BD, Gubler DB, Fitzgerald LA, Rodgers GM. Ho-

mocysteine, a risk factor for premature vascular disease and thrombosis, induces tissue factor activity in endothelial cells. *Arterioscler Thromb* 1993;13:1327-33.

32. Serruys PW, de Jaegere P, Kiemencij F, et al. A comparison of balloon-expandable–stent implantation with balloon angioplasty in patients with coronary artery disease. *N Engl J Med* 1994;331:489-95.

33. Fischman DL, Leon MB, Baim DS, et al. A randomized comparison of coronary-stent placement and balloon angioplasty in the treatment of coronary artery disease. *N Engl J Med* 1994;331:496-501.

34. Robinson K, Arheart K, Refsum H, et al. Low circulation folate and vitamin B6 concentrations: risk factors for stroke, peripheral vascular disease, and coronary artery disease. *Circulation* 1998;97:437-43. [Erratum, *Circulation* 1999;99:983.]

35. Krishnamurthi S, Kakkar VV. Studies on the effect of platelet inhibitors on platelet adhesion to collagen and collagen-induced human platelet activation. *Thromb Haemost* 1985;53:337-42.

Copyright © 2001 Massachusetts Medical Society.